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Efficacy of a yeast derivative on broiler performance, intestinal morphology and blood profile

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ABSTRACT

The trial objective was to investigate the efficacy of a yeast derivative (YD) on performance, health status, distal jejunal structure, and blood profile of broiler chickens.
In a 35 day feeding trial, 1 day-old broilers were allocated to 3 experimental groups with 8 replicates (26/pen): control, treatment 1 (1 kg YD/t feed) and treatment 2 (2 kg YD/t feed). Feed and water were provided ad libitum. On day 35 the distal jejunum was collected from 8 chickens per group (1/pen) for morphometric measurements. Blood samples were collected (1/pen) on day 35 and analyzed by flow cytometry for leukocyte, heterophil, lymphocyte and monocyte enumeration.
In the course of the trial a positive influence was observed by supplementation of YD. On day 35 body weight (BW) (1520 g) and daily weight gain (42.37 g/day) were greater in birds receiving 1 kg/t of YD compared to the control (1374 g BW, 38.18 g/day, respectively; $P=0.05$). Supplementation with 2 kg/t of YD did not improve 35 day BW but did improve cumulative feed-to-gain ratio by 13% ($P<0.05$) versus birds fed the control diet. Histological evaluation demonstrated greater goblet cell density in birds fed either concentration of YD ($P<0.05$). Villus height and crypt depth, however, were unaffected. Apoptotic enterocytes were decreased by both concentrations of YD ($P=0.02$). Supplementation of YD had no effect on blood cell counts ($P>0.05$). The inclusion of YD in diets of broilers was able to improve broiler performance, intestinal goblet cell numbers and to reduce numbers of enterocytes undergoing apoptosis.

1. Introduction

Yeast products have been fed to animals for more than a hundred years, either in the form of yeast fermented mash produced on the farm, yeast by-products from breweries and distilleries, or yeast products commercially produced for animal feeding. However, their *in vitro* and *in vivo modes* of action, in improving animal performance are still not well understood. Live yeasts have been described as being capable of protecting the intestinal mucosa against invading microorganisms by being antagonistic to undesirable microorganisms (barrier effect), and by contributing to the stimulation and maturation of the host animal's immune response (Fuller, 1989). Yeast cell wall components (e.g. mannanoligosaccharides) reportedly prevent colonization of pathogens in the intestinal tract by binding of pathogenic bacteria which have mannose-specific type-I fimbriae and by having prebiotic activity, which leads to an increase in number of beneficial bacteria such as lactobacilli and bifidobacteria (Firon et al., 1984; Gibson and Wang, 1994; Newman, 1994; Ofek and Beachey, 1978; Shoaf-Sweeney and Hutkins, 2008; Zopf and Roth, 1996). Furthermore, mannanoligosaccharides have been used due to their purported intestinal health benefits. Baurhoo et al. (2007 and 2009)

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and Solis de los Santos et al. (2007) investigated mannanoligosaccharide preparations in chickens and observed improved gut morphology including longer villi, shorter crypts and enhanced goblet cell numbers in the villus. Beyond the surface area improvement, goblet cell numbers are critical due to production of intestinal mucus. Mucus has several functions such as lubricating intestinal surfaces, trapping and neutralizing bacteria, detoxifying heavy metals by binding, interacting with the intestinal immune system, acting as a diffuse barrier for nutrients and macromolecules thus protecting the underlying epithelial cells (Forstner and Forstner, 1994). Another important function of mucin is that the structure of the mucus provides several potential attachment sites and colonization niches for commensal and pathogenic bacteria (Sonnenburg et al., 2004).

Furthermore, yeast-derived polysaccharides, such as β-glucans, have been described to have the ability to modulate the immune system. The most active immune stimulators appear to be branch chained 1,3-β-D-glucans, sometimes referred to as 1,3- and 1,6- β-D-glucans, present in the yeast cell wall. These branch chain glucans have the capacity to activate the innate immune system, thereby enhancing the defense barriers and providing protection against a variety of infections (Raa, 1996; Stuyven et al., 2009). Heterophils and monocytes, which belong to the innate immune system, have the ability to phagocyte bacteria and parasites to protect the animal against primary infections. The heterophil is a highly phagocytic, granulated cell capable of antimicrobial activity and is analogous, although not identical, to the mammalian neutrophil (Harmon, 1998; Huff et al., 2007). Huff et al. (2007) reported an increase in heterophil (percentage of leucocytes) in whole blood in a study in which turkeys were fed yeast extract and challenged with *E. coli*. While activation of heterophils is an important component of the innate immune response, heterophils are also responsible for tissue damage by accumulation in inflamed tissue and forming heterophil granulomas that are morphologically similar to inflammatory lesions in reptiles (Harmon, 1998). In mammals, stress and infection have been shown to increase the rate of neutrophil production by 10-fold (Smith, 1994). Numerous studies, summarized by Kogan and Kocher (2007), have shown that yeast derivatives, such as β-glucan fractions, enhance the functional status of macrophages; however, little is known about the effect of yeast derivatives on the number of monocytes, the precursor of macrophages.

The objective of the present study was to investigate a YD for its ability to influence broiler performance, intestinal characteristics (morphology, goblet cell numbers, and numbers of apoptotic enterocytes), and blood cell profile (monocytes, heterophils and lymphocytes).

2. Material and methods

2.1. Experimental design

Day-old Ross 308 broiler chicks (mixed sex) were randomly assigned to one of three dietary treatments for the 35 day feeding period. Dietary treatments included feeding YD at 0, 1, or 2 kg/t diet to 8 replicate pens of 26 chicks per pen. Each pen was 1.32 m^2 and had clean wood shavings litter, nipple drinkers and automatic feeders. Climate conditions and the lighting program were controlled by a computer system according to the standard recommendations for broiler chicks, and the changes in environmental conditions were automatically recorded daily. The diet formulation and calculated nutrient composition are shown in Table 1.

2.2. Yeast derivative

The yeast preparation used in the study contains yeast cell wall fragments and yeast extract derived from *Saccharomyces cerevisiae*. The cell wall fragments are obtained by centrifugation from an autolysed yeast culture. The pellet containing the yeast cell wall fragments and partially also the supernatant is than spray dried. The YD was analyzed for mannan-, glucan- and crude protein contents and the capacity to bind *Salmonella typhimurium* and *E. coli* F4 *in vitro*. Total mannan and glucan were determined by HPLC using an isocratic method and a Refractive Index (RI) Detector. Polysaccharides were hydrolyzed with 72% sulfuric acid; with a subsequent Carrez-precipitation interfering materials were removed. Using the Carrez-precipitation distracting proteins and fats are precipitated as zinc (2+) and/or cyanoferrat (II)-complexes (Matissek et al., 1992). Total protein was determined by Total Kjeldahl Nitrogen (TKN, AOAC International Method 954.01).

The *in vitro* capacity to bind *S. typhimurium* and *E. coli* F4 was determined with a quantitative microbiological

Table 1
Basal diet formulation of the starter and grower diets.

Ingredients %	Diets	
	Starter diet (0 to 14 days of age)	Grower diet (14 to 35 days of age)
Raw materials, g/kg		
Corn (maize)	579.3	597.5
Soybean meal, 48% CP	312.5	296.0
Vitamin–mineral premix	62.5[a]	60.0[b]
Soy oil	25.0	20.0
Vegetable oil blend	12.5	25.0
L-Lysine	3.8	1.5
Monocalcium phosphate	2.5	–
L-Threonine	1.3	–
DL-Methionine	0.8	–
Nutrient composition		
kJ/kg	12,648	12,958
Crude protein, g/kg	208.1	197.4
Methionine, g/kg	5.49	4.59
Met + Cys, g/kg	9.02	8.02
Lysine, g/kg	14.06	11.78
Threonine, g/kg	9.27	7.77
Calcium, g/kg	11.96	11.10
Phosphorus, g/kg	8.47	7.68

[a] Supplied per kg of feed: vitamin A 14,450 I.U.; vitamin D_3, 5000 I.U.; vitamin E, 100 mg; vitamin K_3, 3,6 mg; vitamin C, 80 mg; vitamin B_1, 3 mg; vitamin B_2, 6 mg; vitamin B_6, 6 mg; vitamin B_{12}, 40 μg; nicotinic acid, 90 mg; Ca-pantothenic acid, 16 mg; cholinchlorid, 525 mg; folic acid, 2 mg; biotin, 270 μg; copper, 25 mg; zinc 75 mg; iron 125 mg; manganese, 75 mg; cobalt, 1.25 mg; iodine, 2.5 mg; selenium, 0.5 mg.

[b] Supplied per kg of feed: vitamin A 13,850 I.U.; vitamin D_3, 4800 I.U.; vitamin E, 95 mg; vitamin K_3, 3.4 mg; vitamin C, 80 mg; vitamin B_1, 3 mg; vitamin B_2, 8.5 mg; vitamin B_6, 6 mg; vitamin B_{12}, 40 μg; nicotinic acid, 85 mg; Ca-pantothenic acid, 15 mg; cholinchlorid, 500 mg; folic acid, 2 mg; biotin, 260 μg; copper, 24 mg; zinc 72 mg; iron 120 mg; manganese, 72 mg; cobalt, 1.2 mg; iodine, 2.4 mg; selenium, 0.5 mg.